

Data Evaluation Report on the Acute Toxicity of Gamma Cyhalothrin, XDE-225 and Lambda-Cyhalothrin, to Freshwater Invertebrates - *Daphnia magna*

PMRA Submission Number{.....}

EPA MRID Number 45447220

| | | |
|--------------------------|-----------------|---------------|
| Data Requirement: | PMRA DATA CODE | {.....} |
| | EPA DP Barcode | Not available |
| | OECD Data Point | |
| | EPA MRID | 45447220 |
| | EPA Guideline | 72-2 |

Test material: Gamma Cyhalothrin

Common name: XDE-225

Purity: 100%

Chemical name: IUPAC : Not reported

CAS name: Not reported

CAS No.: 76703-62-3

Synonyms: Not reported

Common name: Lambda-Cyhalothrin isomer

Purity: 99%

Chemical name: IUPAC : Not reported

CAS name: Not reported

CAS No.: 91465-08-06

Synonyms: Not reported

Primary Reviewer: Rebecca Bryan
Staff Scientist, Dynamac Corporation**Signature:** *Rebecca Bryan*
Date: 6/30/03**QC Reviewer:** Dana Worcester
Staff Scientist, Dynamac Corporation**Signature:** *Dana Worcester*
Date: 6/30/03**Primary Reviewer:** *Norman Birchfield*
OPP/EFED/ERB|**Date:** *Norman Birchfield*
*1/26/2004***Secondary Reviewer(s):**
{EPA/OECD/PMRA}**Date:****Reference/Submission No.:****Company Code:****Active Code:****EPA PC Code:** 128807**Date Evaluation Completed:**

CITATION: Machado, M.W. 2001. XDE-225 and Lambda-Cyhalothrin: Comparative Toxicity to Daphnids (*Daphnia magna*) under Static-Renewal Conditions. Unpublished study performed by Springborn Laboratories, Inc., Wareham, Massachusetts. Springborn Project ID No. 12550.6139. Study submitted by The Dow Chemical Company, Midland, Michigan. Experimental start date April 17, 2001 and experimental termination date April 19, 2001. Final report issued May 21, 2001.



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The 48-hour acute toxicity of gamma cyhalothrin (XDE-225) and lambda cyhalothrin to the water flea, *Daphnia magna*, was studied under static renewal conditions.

XDE-225: Daphnids were exposed to the test material at nominal concentrations of 0 (negative and solvent controls), 0.031, 0.063, 0.13, 0.25, and 0.50 µg a.i./L; mean-measured concentrations were <0.0025-0.0026 (LOQ, negative and solvent controls), 0.015, 0.034, 0.060, 0.14, and 0.25 µg a.i./L. After 48 hours, there was 25, 55, 50, 75, and 75% immobility in the 0.015, 0.034, 0.060, 0.14, and 0.25 µg a.i./L XDE-225 treatment groups. After 48 hours, all mobile daphnids were observed to be lethargic, on the bottom of test vessels, and swimming, carrying particulate matter. No immobility was observed in the dilution water or solvent controls. The 48-hour EC₅₀ (with 95% C.I.) was 0.045 (0.020-0.079) µg a.i./L, which categorizes Gamma Cyhalothrin (XDE-225) as very highly toxic to the water flea (*Daphnia magna*) on an acute toxicity basis. The 48-hour NOEC level was <0.015 µg a.i./L, the lowest concentration tested.

Lambda-Cyhalothrin: Daphnids were exposed to the test material at nominal concentrations of 0 (negative and solvent controls), 0.013, 0.025, 0.050, 0.10, and 0.20 µg a.i./L; mean-measured concentrations were <0.0025-0.0026 (LOQ, negative and solvent controls), 0.055, 0.012, 0.023, 0.050, and 0.079 µg a.i./L. After 48 hours, there was 5, 20, 30, 30, and 75% immobility in the 0.0055, 0.012, 0.023, 0.050, and 0.079 µg a.i./L Lambda-Cyhalothrin treatment groups. No immobility was observed in the dilution water or solvent controls. After 48 hours, all mobile daphnids were observed to be lethargic, on the bottom of test vessels, and swimming, carrying particulate matter. The 48-hour EC₅₀ (with 95% C.I.) was 0.051 (0.034-0.10) µg a.i./L, which categorizes Lambda-Cyhalothrin as very highly toxic to the water flea (*Daphnia magna*) on an acute toxicity basis. The 48-hour NOEC level was <0.0055 µg a.i./L, the lowest concentration tested.

This comparison study is scientifically sound and satisfies the guideline requirements for an acute toxicity study with freshwater invertebrates (§72-2). This study is classified as CORE.

Results Synopsis

Test Organism Age (eg. 1st instar): <24 hours old

Test Type (Flow-through, Static, Static Renewal): Static Renewal

48-Hour

XDE-225:

EC₅₀: 0.045 µg a.i./L 95% C.I.: 0.020-0.079 µg a.i./L

Slope: 1.08 (0.47-1.69)

NOEC: <0.015 µg a.i./L

LOEC: 0.015 µg a.i./L

Lambda-Cyhalothrin:

EC₅₀: 0.051 µg a.i./L 95% C.I.: 0.034-0.10 µg a.i./L

Slope: 1.61 (0.87-2.35)

NOEC: <0.0055 µg a.i./L

LOEC: 0.0055 µg a.i./L

I. MATERIALS AND METHODS

GUIDELINE FOLLOWED: The study protocol was based on procedures outlined in Ecological Effects Test Guideline OPPTS 850.1010 Aquatic Invertebrate Acute Toxicity Test, Freshwater Daphnids (U.S. EPA, 1996); and OECD guideline No. 202 for acute testing on freshwater invertebrates. Deviations from §72-2 included:

1. The loading rate was not specified.
2. The hardness (170 mg/L as CaCO_3) was higher than recommended (40-48 mg/L as CaCO_3). The pH (8.0-8.1) was greater than recommended (7.2-7.6).

These deviations did not affect the acceptability or the validity of the study.

COMPLIANCE: Signed and dated GLP, Confidentiality, and Quality Assurance statements were provided.

A. MATERIALS:

1a. Test Material XDE-225

Description: Not reported

Lot No./Batch No. : CH519JMi87ABLSE

Purity: 100%

Stability of Compound

Under Test Conditions: For XDE-225, mean analyzed concentrations were 62.9-82.0% of nominal concentrations for "new" solutions (days 0 and 1) and 27.7-38.1% for "old" solutions (days 1 and 2) (Table 2, p. 28).

OECD requires water solubility, stability in water and light, pK_a , P_{ow} and vapor pressure of the test compound. OECD requirements were not reported.

**Storage conditions
of test chemical:**

Stored at room temperature in the dark.

1b. Test Material Lambda -Cyhalothrin

Description: Not reported

Lot No./Batch No. : CHA DENMARK 1-1

Purity: 99%

Stability of Compound

Under Test Conditions: For Lambda-Cyhalothrin, mean analyzed concentrations were 52.3-70.0% of nominal concentrations for "new" solutions (days 0 and 1) and 26.3-34.5% for "old" solutions (days 1 and 2)(Table 6, p. 32).

OECD requires water solubility, stability in water and light, pK_a , P_{ow} and vapor pressure of the test compound. OECD requirements were not reported.

Storage conditions of test chemical:

Stored at room temperature in the dark.

2. Test organism:

Species: *Daphnia magna*

Age at test initiation: ≤ 24 hours old

Source: In-house laboratory cultures.

B. STUDY DESIGN:

1. Experimental Conditions

a) Range-finding Studies:

XDE-225: Definitive test concentrations were based upon results of a range-finding test. A static-renewal preliminary test was conducted at nominal concentrations of 0.010, 0.050, 0.10, 0.50, and 1.0 $\mu\text{g a.i./L}$ with dilution water control. There was 30, 20, and 90% immobilization for the 0.050, 0.10, and 0.50 $\mu\text{g a.i./L}$ treatment groups. Lethargy was observed in the remaining daphnids of all the treatment groups. The control and 0.010 $\mu\text{g a.i./L}$ treatment group had no immobilization.

Lambda-Cyhalothrin: Definitive test concentrations were based upon results of a range-finding test. A static-renewal preliminary test was conducted at nominal concentrations of 0.030, 0.15, 0.30, 1.5, and 3.0 $\mu\text{g a.i./L}$ with dilution water control. There was 50, 40, 50, 70, and 90% immobilization for the 0.030, 0.15, 0.30, 1.5, and 3.0 $\mu\text{g a.i./L}$ treatment groups. Lethargy was observed in the daphnids of all the treatment groups. The control had no immobilization.

b) Definitive Study

Table 1 . Experimental Parameters

| Parameter | Details | Remarks |
|---|---|--|
| | | Criteria |
| Acclimation period: | Continuous laboratory cultures were maintained. | Daphnids were not fed during the test. |
| Conditions: (same as test or not) | Same as test | Both XDE-225 and Lambda-Cyhalothrin tests. |
| Feeding: | <i>Daphnia</i> cultures were fed 2 mL/vessel/day of <i>Ankistrodesmus falcatus</i> (4 x 10 ⁷ cells/mL) and 0.5 mL of YCT trout chow (yeast, cereal leaves, and flaked fish food), daily. | EPA requires 7 day minimum acclimation period. No feeding during study. |
| Health: (any mortality observed) | Successfully survived and reproduced over several generations. | |
| Duration of the test | 48 hours | Both XDE-225 and Lambda-Cyhalothrin tests. |
| | | EPA requires 48 hours |
| Test condition - static/flow through | Static renewal | Both XDE-225 and Lambda-Cyhalothrin tests. |
| Type of dilution system (for flow through method) | N/A | |
| Renewal rate (for static renewal) | Day 1 | EPA requires consistent flow rate of 5 - 10 volumes/24 hours, meter systems calibrated before study and checked twice daily during test period |
| Aeration, if any | N/A | Both XDE-225 and Lambda-Cyhalothrin tests. |
| | | |
| <u>Test vessel</u> | | Both XDE-225 and Lambda-Cyhalothrin tests. |
| Material: (glass/stainless steel) | Glass beakers | |
| Size: | 1000 mL | EPA requires: size 20 ml or 3.9 L |
| Fill volume: | 800 mL | fill 200 ml |

| Parameter | Details | Remarks |
|--------------------------|--|--|
| | | Criteria |
| Source of dilution water | The dilution water was fortified well water that was filtered through an Amberlite XAD-7 resin column. | Both XDE-225 and Lambda-Cyhalothrin tests. <i>EPA requires soft reconstituted water or water from a natural source, not dechlorinated tap water.</i> |
| <u>Water parameters:</u> | | |
| Hardness | 170 mg/L as CaCO ₃ (Both XDE-225 and Lambda-Cyhalothrin tests). | The hardness (170 mg/L as CaCO ₃) was higher than recommended (40-48 mg/L as CaCO ₃). The pH (8.0-8.1) was greater than recommended (7.2-7.6). |
| pH | <u>XDE-225</u> : 8.0-8.1 <u>Lambda-Cyhalothrin</u> : 8.0-8.1 | |
| Dissolved oxygen | <u>XDE-225</u> : 8.3-8.9 (≥93%) <u>Lambda-Cyhalothrin</u> : 8.4-8.9 (≥94%) | |
| Temperature | 21-22°C (Both XDE-225 and Lambda-Cyhalothrin tests). | |
| Total Organic Carbon | 0.58 mg/L (Both XDE-225 and Lambda-Cyhalothrin tests). | |
| Particulate matter | Not reported | <i>EPA requires:</i> <i>hardness: 40 - 48 mg/L as CaCO₃</i> <i>pH: 7.2 - 7.6</i> <i>-Temperature: 20°C (measured continuously or if water baths are used, every 6 hr, may not vary > 1°C</i> <i>Dissolved oxygen:</i> <i>Static: ≥ 60% during 1st 24 hr and ≥ 40% during 2nd 24 hr</i> <i>Flow-through: ≥60%</i> |
| Metals | Not at toxic concentrations (Both XDE-225 and Lambda-Cyhalothrin tests). | |
| Pesticides | Not at toxic concentrations (Both XDE-225 and Lambda-Cyhalothrin tests). | |
| Chlorine | Not reported | |

| Parameter | Details | Remarks |
|-----------------------------------|---------|---|
| | | Criteria |
| Number of replicates | | Both XDE-225 and Lambda-Cyhalothrin tests. |
| Solvent control: | 2 | |
| Negative control: | 2 | |
| Treatments: | 2 | |
| Number of organisms per replicate | | Both XDE-225 and Lambda-Cyhalothrin tests. |
| Solvent control: | 10 | |
| Negative control: | 10 | |
| Treatments: | 10 | The biomass loading rate was not specified. |
| | | EPA requires 5 treatment levels plus control with a minimum of 20 daphnid per treatment. Biomass loading rate for static ≤ 0.8 g/L at $\leq 17^{\circ}\text{C}$, ≤ 0.5 g/L at $> 17^{\circ}\text{C}$; flow-through: ≤ 1 g/L/day. |

| Parameter | Details | Remarks |
|---|--|---|
| | | Criteria |
| Treatment concentrations nominal: measured: | <p><u>XDE-225</u>: 0 (negative and solvent controls), 0.031, 0.063, 0.13, 0.25, and 0.50 µg a.i./L</p> <p><u>Lambda-Cyhalothrin</u>: 0 (negative and solvent controls), 0.013, 0.025, 0.050, 0.10, and 0.20 µg a.i./L</p> <p><u>XDE-225</u>: <0.0025-0.0026 (LOQ, negative and solvent controls), 0.015, 0.034, 0.060, 0.14, and 0.25 µg a.i./L</p> <p><u>Lambda-Cyhalothrin</u>: <0.0025-0.0026 (LOQ, negative and solvent controls), 0.055, 0.012, 0.023, 0.050, and 0.079 µg a.i./L</p> | |
| | | <i>EPA requires a geometric series with each concentration being at least 60% of the next higher one.</i> |
| Solvent (type, percentage, if used) | Acetone, 0.1 mL/L | Both XDE-225 and Lambda-Cyhalothrin tests. |
| | | <i>EPA requires solvents not to exceed 0.5 mL/L for static tests or 0.1 mL/L for flow-through tests.</i> |
| Lighting | 16 hours light/8 hours dark with a transition period. | Both XDE-225 and Lambda-Cyhalothrin tests. |
| | | <i>EPA requires 16 hours light, 8 hours dark.</i> |

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| Parameter | Details | Remarks |
|--|--|--|
| | | Criteria |
| Stability of chemical in the test system | <p><u>XDE-225</u>: Mean analyzed concentrations were 62.9-82.0% of nominal concentrations for "new" solutions (days 0 and 1) and 27.7-38.1% for "old" solutions (days 1 and 2).</p> <p><u>Lambda-Cyhalothrin</u>: Mean analyzed concentrations were 52.3-70.0% of nominal concentrations for "new" solutions (days 0 and 1) and 26.3-34.5% for "old" solutions (days 1 and 2).</p> | The average "old" and "new" test solutions were reviewer-calculated from measured concentrations. |
| Recovery of chemical | <p><u>XDE-225</u>: Mean measured concentrations were 46-57% of nominal.</p> <p><u>Lambda-Cyhalothrin</u>: Mean measured concentrations were 39-50% of nominal.</p> | Recoveries were based on average of "new" solutions (days 0 and 1) and "old" solutions (days 1 and 2) (Table 2, p. 28 and Table 6, p. 32). |
| Level of Quantitation | 0.0025-0.0026 µg a.i./L(Both XDE-225 and Lambda-Cyhalothrin tests.) | |
| Level of Detection | Not reported | |
| Positive control {if used, indicate the chemical and concentrations} | N/A | |
| Other parameters, if any | N/A | |

2. Observations:

Table 2: Observations

| Criteria | Details | Remarks |
|---|---|--|
| | | Criteria |
| Parameters measured including the sublethal effects | Mortality/immobility Other sublethal effects | Both XDE-225 and Lambda-Cyhalothrin tests. |
| Observation intervals | After 24 and 48 hours | Both XDE-225 and Lambda-Cyhalothrin tests. |
| Were raw data included? | Yes, sufficient | Both XDE-225 and Lambda-Cyhalothrin tests. |
| Other observations, if any | N/A | |

II. RESULTS AND DISCUSSION

A. SUB-LETHAL TOXICITY ENDPOINTS:

Table 3a: Sublethal Effect of XDE-225 on *Daphnia magna*.

| Treatment µg a.i./L (nominal conc.) | Observation period | | | |
|---|--------------------|------------|---------------------|------------|
| | 24 hours | | 48 hours | |
| | endpoint | % affected | endpoint | % affected |
| Dilution water Control | Immobile | 0 | Immobile | 0 |
| Solvent Control | Immobile | 0 | Immobile | 0 |
| Positive control, if used | N/A | N/A | N/A | N/A |
| 0.015 (0.031) | Immobile | 0 | Immobile | 25 |
| 0.034 (0.063) | Immobile | 5 | Immobile | 55 |
| 0.060 (0.13) | Immobile | 0 | Immobile | 50 |
| 0.14 (0.25) | Immobile | 0 | Immobile | 75 |
| 0.25 (0.50) | Immobile | 5 | Immobile | 75 |
| NOEC, µg a.i./L | Not specified | | <0.015 | |
| LOEC, µg a.i./L | Not specified | | 0.015 | |
| EC ₅₀ (with 95% C.I.), µg a.i./L | >0.25 | | 0.045 (0.020-0.079) | |

Table 3b: Sublethal Effect of Lambda-Cyhalothrin on *Daphnia magna*.

| Treatment µg a.i./L (nominal conc.) | Observation period | | | |
|---|--------------------|------------|--------------------|------------|
| | 24 hours | | 48 hours | |
| | endpoint | % affected | endpoint | % affected |
| Dilution water Control | Immobile | 0 | Immobile | 0 |
| Solvent Control | Immobile | 0 | Immobile | 0 |
| Positive control, if used | N/A | N/A | N/A | N/A |
| 0.0055 (0.013) | Immobile | 0 | Immobile | 5 |
| 0.012 (0.025) | Immobile | 0 | Immobile | 20 |
| 0.023 (0.050) | Immobile | 0 | Immobile | 30 |
| 0.050 (0.10) | Immobile | 0 | Immobile | 30 |
| 0.079 (0.20) | Immobile | 0 | Immobile | 75 |
| NOEC, µg a.i./L | Not specified | | <0.0055 | |
| LOEC, µg a.i./L | Not specified | | 0.0055 | |
| EC ₅₀ (with 95% C.I.), µg a.i./L | >0.079 | | 0.051 (0.034-0.10) | |

There was 25, 55, 50, 75, and 75% immobility in the 0.015, 0.034, 0.060, 0.14, and 0.25 µg a.i./L XDE-225 treatment groups. There was 5, 20, 30, 30, and 75% immobility in the 0.0055, 0.012, 0.023, 0.050, and 0.079 µg a.i./L Lambda-Cyhalothrin treatment groups. After 48 hours, all mobile daphnids in the XDE-225 and Lambda-Cyhalothrin treatment groups were observed to be lethargic, on the bottom of test vessels, and swimming, carrying particulate matter.

B. REPORTED STATISTICS:

Statistical Method: The EC₅₀ values (with 95% C.I.) after 48 hours were calculated using probit analysis (Stephan computer program). The NOEC was visually determined based on immobility data (Both XDE-225 and Lambda-Cyhalothrin tests).

C. VERIFICATION OF STATISTICAL RESULTS:

Statistical Method: The LC₅₀ for XDE-225 and Lambda-Cyhalothrin were determined using the US EPA Probit method via TOXANAL statistical software. The NOEC for both compounds was visually determined based on sublethal effects.

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48-Hour

EC₅₀: 0.045 µg a.i./L 95% C.I.: 0.020-0.079 µg a.i./L

Slope: 1.08 (0.47-1.69)

NOEC: <0.015 µg a.i./L

LOEC: 0.015 µg a.i./L

Lambda-Cyhalothrin:

EC₅₀: 0.051 µg a.i./L 95% C.I.: 0.034-0.10 µg a.i./L

Slope: 1.61 (0.87-2.35)

NOEC: <0.0055 µg a.i./L

LOEC: 0.0055 µg a.i./L

D. STUDY DEFICIENCIES:

There were no significant deviations from U.S. EPA guideline §72-2a that affected the acceptability of this study.

E. REVIEWER'S COMMENTS:

The reviewer's conclusions were identical to the study author's. The 48-hour EC₅₀ values for XDE-225 and Lambda-Cyhalothrin were 0.045 and 0.051 µg a.i./L, which categorizes these compounds as very highly toxic to the water flea (*Daphnia magna*) on an acute toxicity basis.

For XDE-225, analyzed concentrations were 62.9-82.0% of nominal concentrations for "new" solutions (days 0 and 1) and 27.7-38.1% for "old" solutions (days 1 and 2). For Lambda-Cyhalothrin, analyzed concentrations were 52.3-70.0% of nominal concentrations for "new" solutions (days 0 and 1) and 26.3-34.5% for "old" solutions (days 1 and 2).

The study author reported that the aquatic residues for XDE-225 and Lambda-Cyhalothrin decreased between new and aged solutions during the 24-hour renewal period. However, the concentration gradients were maintained.

A flow-through system may have provided better recoveries.

G. CONCLUSIONS:

This comparison study is scientifically sound, fulfills U.S. EPA guideline §72-2, and is classified as CORE. Based on the results of these studies, XDE-225 and Lambda-Cyhalothrin are categorized as very highly toxic to the water flea, *Daphnia magna*, on an acute toxicity basis.

48-Hour

XDE-225:

EC₅₀: 0.045 µg a.i./L 95% C.I.: 0.020-0.079 µg a.i./L

Slope: 1.08 (0.47-1.69)

NOEC: <0.015 µg a.i./L

LOEC: 0.015 µg a.i./L

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Lambda-Cyhalothrin:

EC₅₀: 0.051 µg a.i./L 95% C.I.: 0.034-0.10 µg a.i./L

Slope: 1.61 (0.87-2.35)

NOEC: <0.0055 µg a.i./L

LOEC: 0.0055 µg a.i./L

III. REFERENCES:

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APPENDIX 1. OUTPUT OF REVIEWER'S STATISTICAL VERIFICATION:

XDE-225

RESULTS CALCULATED USING THE MOVING AVERAGE METHOD

| SPAN | G | LC50 | 95 PERCENT CONFIDENCE LIMITS |
|--------------|----------|--------------|------------------------------|
| 4 | .3241892 | 4.354639E-02 | 1.688405E-02 |
| 7.729754E-02 | | | |

RESULTS CALCULATED USING THE PROBIT METHOD

| ITERATIONS | G | H | GOODNESS OF FIT PROBABILITY |
|------------|----------|---|-----------------------------|
| 2 | .3188672 | 1 | .6265266 |

SLOPE = 1.07806

95 PERCENT CONFIDENCE LIMITS = .4692977 AND 1.686823

LC50 = 4.498318E-02

95 PERCENT CONFIDENCE LIMITS = 1.920299E-02 AND .0793904

LC10 = 2.985441E-03

95 PERCENT CONFIDENCE LIMITS = 5.481252E-05 AND 9.664756E-03

Lambda cyhalothrin

RESULTS CALCULATED USING THE MOVING AVERAGE METHOD

| SPAN | G | LC50 | 95 PERCENT CONFIDENCE LIMITS |
|----------|---------|--------------|------------------------------|
| 2 | .474772 | 5.839953E-02 | 3.986557E-02 |
| .1308515 | | | |

RESULTS CALCULATED USING THE PROBIT METHOD

| ITERATIONS | G | H | GOODNESS OF FIT PROBABILITY |
|------------|----------|---|-----------------------------|
| 3 | .2112256 | 1 | .1874602 |

SLOPE = 1.610308

95 PERCENT CONFIDENCE LIMITS = .8702218 AND 2.350394

LC50 = 5.164681E-02

95 PERCENT CONFIDENCE LIMITS = 3.451467E-02 AND .1043741

LC10 = 8.401842E-03

95 PERCENT CONFIDENCE LIMITS = 2.549089E-03 AND 1.414095E-02